A biochemical study of Huntington's chorea

F. E. KENYON AND S. M. HARDY

From the Maudsley and Dulwich Hospitals, London

It is now generally accepted that Huntington's chorea is inherited through a single dominant autosomal gene but apart from this little is known of the aetiology. Earlier investigators had implicated endocrine factors (Mulon and Porak, 1912; Bize, 1934) but without very convincing evidence. With the advent of new drugs that had profound effects on the nervous system the idea of a neuronal basis for Huntington's chorea received a fresh impetus but a recent review showed little general agreement (Walter-Buel, 1956). More recently other workers have suggested that the condition could be regarded as a distorted form of psychomotility (Kempinsky, Boniface, Morgan, and Busch, 1960). A recent review of the pathology, after showing that typical changes in the basal ganglia together with non-specific cortical changes are constant features of the condition, ends by suggesting an enzyme deficiency as the responsible factor (McCaughey, 1961). The recent spectacular advances in biochemical genetics had previously led others to speculate on a possible enzymatic aetiology (*e.g.*, Penrose, 1958) but as yet there has been no convincing proof.

There are relatively few biochemical studies of Huntington's chorea reported in the literature and in most cases they deal with very small numbers (Falstein and Stone, 1940; Nielsen and Butt, 1955; Forrest, 1957; Chhuttani, Chopra, and Singh, 1959; Oliphant, Evans, and Forrest, 1960; Barbeau, Murphy, and Sourkes, 1961; Williams, Maury, and Kibler, 1961; Perry, 1961). None of these studies has produced any consistently positive findings.

Recently there has been a recrudescence of interest in magnesium metabolism and it occurred to one of us (F.E.K.) that it would be worthwhile investigating this in Huntington's chorea, mainly because choreoathetosis and other types of involuntary movements, as well as a variety of abnormal mental states, have been described in patients suffering from disordered magnesium metabolism (*British Medical Journal*, 1960; MacIntyre, 1960; *Lancet*, 1960; Hanna, 1961; Wallach, Cahill, Rogan, and Jones, 1962) and on a possible analogy with manganese, which is known to damage the basal ganglia (Cotzias, 1958) but has been shown to be normal in Huntington's chorea (Perry, 1961).

MATERIAL AND METHOD

Twenty patients with Huntington's chorea were studied, 16 of them being in mental hospitals at the time. There were 12 females and eight males, with a mean age of 51 years and a mean length of history of 8·6 years. As far as could be ascertained from the case records the mean age at onset was 42·3 years (range 20 to 66 years). There was a positive family history in 14, negative in four, and insufficient information in two. For comparison a control group of 20 schizophrenic patients admitted to an observation ward was similarly studied. They had a mean age of 31·1 years and a mean length of history of 4·7 years.

Blood was taken from both groups by venepuncture, two samples in most cases being obtained, one clotted and one heparinized. The clotted blood was centrifuged and the serum used for magnesium and calcium estimations. Packed cell volume was measured on the heparinized blood in order to correct the magnesium results of the erythrocytes. Then an aliquot amount of heparinized and well-mixed sample of blood was washed three times with physiological saline. The washed erythrocytes were then separated and made up with distilled water to volume.

The protein was precipitated with trichloracetic acid and the magnesium content of the erythrocytes estimated in the supernatant by the method of Szmuk-Fekete with E.D.T.A. titration using erichrome black-T indicator, which indicates both the magnesium and calcium chelation (Balint and Hegedüs, 1955). From this was deducted the result of another E.D.T.A. titration using calcein thymolphthalein indicator, which is our modification of the method of Baron and Bell (1957).

RESULTS

The mean values of serum and cell magnesium and calcium for patients with Huntington's chorea compared with those in the control group are given in Table I. Results are shown separately for two subgroups of Huntington's chorea, namely, those with and those without a known family history of the condition, to see whether there is any biochemical support for this subdivision, as has recently been suggested on genealogical grounds (Lyon, 1962). From Table I it can be seen that there is no significant difference between any of the groups for serum magnesium and calcium. The main differences found are for the intracellular values; the mean erythrocyte magnesium is higher in the Huntington

F. E. Kenyon and S. M. Hardy

TABLE I

MEAN VALUES FOR MAGNESIUM AND CALCIUM

	Huntington's Chorea		All Huntington's Chorea Cases	Control Group
	Positive Family History	*Negative Family History*		
Erythrocyte magnesium (mEq. per litre)	6·77	13·47	8·87	5·17
Erythrocyte calcium (mEq. per litre)	1·64	1·72	1·55	0·26
Serum magnesium (mEq. per litre)	2·20	2·36	2·14	2·30
Serum calcium (mEq. per litre)	4·65	4·06	4·54	4·60

patients than in controls and in those with a negative compared with a positive family history.

Statistical evaluation showed that in all Huntington patients (N = 15) the mean erythrocyte magnesium value was 8·87 ± 1·89 mEq. per litre, with a range of 2·5 to 25·0, and in schizophrenic controls (N = 15) 5·17 ± 0·43 mEq. per litre, range 4·5 to 7·0. Comparison of these give t = 1·91, which just falls short of the 5% level of significance. For the Huntington subgroups comparable results were: in those with a positive family history (N=10) mean erythrocyte magnesium was 6·77 ± 1·73 mEq. per litre and with a negative family history (N = 4) 13·47 ± 6·01; comparing the two t = 1·07, which is not significant.

In the 13 Huntington patients in whom both serum and cell magnesium were estimated no consistent relationship between the two was found, thus supporting the similar findings of other workers in normal subjects (Wallach *et al.*, 1962). There was no correlation found between length of history and level of erythrocyte magnesium.

Statistical analysis of the erythrocyte calcium values gave the following values:—For all Huntington patients (N = 15) mean calcium was 1·55 ± 0·32 mEq. per litre (range 0 to 2·9); for schizophrenic controls (N = 14) 0·26 ± 0·22 mEq. per litre (range 0 to 3·0) and a comparison of these gives t = 3·31, which is highly significant (P < 0·001).

There was no significant difference in erythrocyte calcium between the subgroups with positive and negative family histories (t = 0·12). As with magnesium there was no consistent relationship between the length of history in subjects with Huntington's chorea and the level of erythrocyte calcium.

Individual results of erythrocyte calcium and magnesium estimations for all the patients, as well as an indication of the family history are given in Table II.

When levels of intracellular calcium and magnesium are compared (Table III), no consistent relationship between the two can be demonstrated. When results from the control group are similarly arranged, again no consistent relationship was noted.

TABLE II

ERYTHROCYTE CALCIUM AND MAGNESIUM VALUES FOR ALL HUNTINGTON PATIENTS

Calcium (mEq. per litre)	Magnesium (mEq. per litre)	Family History
2·9	3·3	+
2·9	3·3	+
2·9	25·0	−
2·5	4·1	+
2·5	4·1	+
2·0	8·3	+
2·0	2·5	−
2·0	19·1	−
2·0	20·8	+
1·6	5·8	+
0	5·3	+
0	6·1	+
0	6·6	+
0	7·3	−
0	11·4	n.k.

TABLE III

COMPARISON OF ERYTHROCYTE CALCIUM AND MAGNESIUM FOR ALL HUNTINGTON PATIENTS

Calcium	Nos.	Magnesium (mean)
>2·0	5	7·96
1-2·0	5	11·30
<1	5	7·34

DISCUSSION

The main findings of this study are the raised intracellular levels of calcium and magnesium in patients with Huntington's chorea. Compared with controls the calcium is much more significantly raised than the magnesium, but magnesium differentiates better those with and those without a family history and also shows a much greater range of variation in the whole group of Huntington patients.

Previous work on calcium metabolism in Huntington's chorea has not produced any consistent findings; Hühnerfeld (1931) found an increased level of calcium in the cerebrospinal fluid in three patient, found a slight hypocalcaemia but the main Bize (1934), in an extensive investigation of one patient, found a slight hypocalcaemia but the main positive result was a high cell potassium level. Other workers after a thorough study of 15 patients report

negative findings for a variety of blood constituents, including calcium (Falstein and Stone, 1940).

There is only one report in the literature known to us in which magnesium estimations in Huntington's chorea are reported, and this concerns the one patient already referred to above, when slightly raised plasma magnesium and slightly lowered cell magnesium levels were found but neither outside the normal range of variation (Bize, 1934). Bize considers that all his different findings could probably be explained by a parathyroid deficiency.

How our own findings can be related to the known neuropathology of Huntington's chorea must at present remain speculative, but the following points make us think that magnesium may be a very important factor.

A recent study on magnesium metabolism stated that 'the human erythrocyte has one fourth the magnesium concentration of other cells' (Wallach et al., 1962); in the light of the present findings this suggests that the neural cells must contain very high concentrations of magnesium in Huntington's chorea.

In a case of experimental manganese poisoning (Grünstein and Popowa, 1929) degeneration of the small cells of the putamen and caudate nucleus was found, changes identical with those reported in Huntington's chorea (McCaughey, 1961). Moreover, manganese and magnesium are to a large extent interchangeable, at least in the activation of various enzyme systems (Cotzias, Borg, and Bertinchamps, 1960).

At one time, but now disproved, it was thought that copper metabolism was at fault in Huntington's chorea and two cases have been reported in which treatment with B.A.L. produced definite clinical improvement (Nielsen and Butt, 1955). However, it is also known that B.A.L., at least in high dosage, can produce a lowering of magnesium levels.

Magnesium has many different functions in the body and the whole pattern of its metabolism is as yet not fully understood but it is known that magnesium is of fundamental importance in energy metabolism, as it forms a complex with adenosine triphosphate and the ratio of the two is critical for certain energy exchange reactions (Mudd, 1959; Hers, 1952; von Szent-Györgyi, 1960; McIlwain, 1962). It is possible that a high cell magnesium might cause the rate of exchange of high energy phosphates to become faster and, if in turn the so-called 'biological clock' is the time unit within which the normal changeover takes place, then this quickened pace might conceivably cause the cortical changes characteristic of senile change which are a feature of the neuropathology of Huntington's chorea.

But in order to establish whether the demonstrated biochemical changes are primary or secondary, further research is necessary into the oxidative phosphorylation mechanism of these patients' erythrocytes and neural cells compared with normals. It would also be of interest to investigate other members of Huntington families and to see whether there is any possibility of identifying by means of erythrocyte calcium and magnesium levels those who are destined to develop the condition. Further, if any satisfactory chelating agent were available that would bind intracellular magnesium and calcium, then a therapeutic trial might prove rewarding. It may be that those patients who are at present diagnosed as suffering from Huntington's chorea are not (biochemically) a homogeneous group, and in the light of the present findings those so diagnosed but without a typical family history need further investigation. However, in this respect it must be noted that our numbers are small and the differences found, although greater for magnesium than for calcium, did not reach statistical significance in either case.

SUMMARY

After a brief review of previous biochemical investigations reasons are given for thinking that magnesium metabolism may be disturbed in Huntington's chorea. Twenty patients with this condition and the same number of schizophrenic controls had estimations performed of serum and erythrocyte magnesium and calcium. Results showed that patients with Huntington's chorea have normal serum values but raised cellular magnesium and calcium values when compared with controls. When patients with a positive family history were compared with those with a negative one differences were also found; those with no family history had a high cell magnesium without a corresponding increase in calcium although numbers were small and results not significant. The implications of these findings are discussed and suggestions made for further research.

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